Deoxygenated Alkylation of Heteroarenes with Alcohols

Undergraduate Research Thesis

Presented in fulfillment of the requirements for a
B.S. with research distinction in chemistry at The Ohio State University

by

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Abstract

The synthesis and functionalization of pharmaceutically useful compounds has long been a major focus in organic/medicinal chemistry. Post-synthetic alkylation of target compounds is a topic of strong interest to the pharmaceutical community. In light of this, the project focus is on designing metal-free conditions to alkylate heteroarenes using readily available alcohols. In the classic Minisci reaction, alcohol addition to heteroarenes results in a remaining alcohol within the product. Alternatively, a gamma radiation-based procedure allows for a spin-center shift mechanism that provides the alcohol free alkylation product. Recently, a photoredox approach using an iridium catalyst has also been developed to carry out this alkylation. The requirement of an iridium photocatalyst and the need for removal of the toxic iridium metal before biological testing, establishes a need for metal-free versions of this transformation. In this project, we have invented a new, alkylation reaction of heteroarenes with alcohols, under metal-free conditions via a peroxide-mediated radical mechanism. Importantly, we have demonstrated that our new method can alkylate isoquinoline, quinoline, 2,6-lutidine and 4-*t*Butylpyridine derivatives without metal catalysts and with a variety of alcohols. In conclusion, our protocol enables access to diverse alkylated drug derivatives under mild conditions, with the promise for divergent functionalizations of heteroarenes with important pharmaceutical applications.

Introduction

Heterocycle is classified as a cyclic compound with more than two types of element as members in its ring. Heterocycles are commonly presented in the pharmaceutical compounds of popular applications. Among these heterocycles, 92% are nitro-containing heterocycles, where N-Heterocycles are considered as the biggest candidate for drugs (Figure 1).

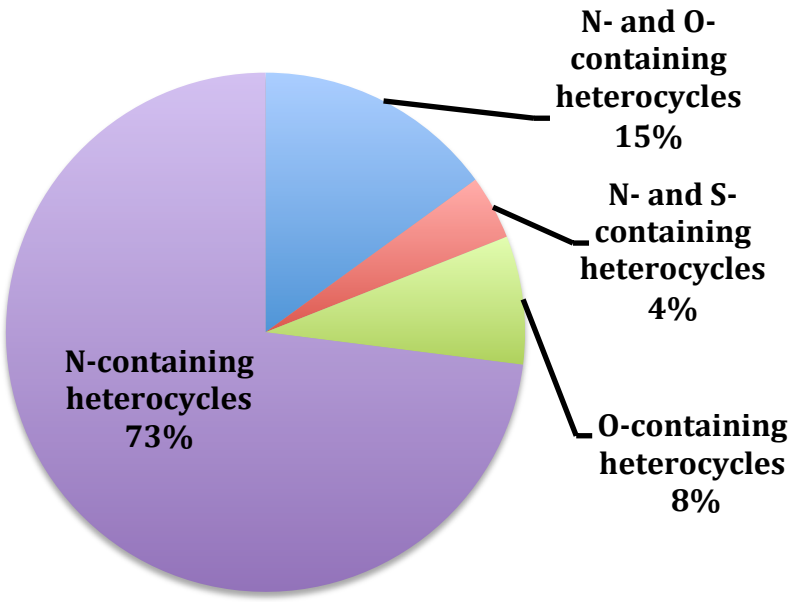


Figure 1 : FDA approved anti-tumor compounds[1] (2010 - 2015)

Heteroarene is the unsaturated version of heterocycle that has aromatic properties. In most pharmaceuticals, heterocycle exists as heteroarene. Among pharmaceutical compounds of heteroarenes derivatives, Berberine, Ibrance, Thiamine and Camptothecin are widely used, and the highlight spots are the interests of alkylation (Figure 2).

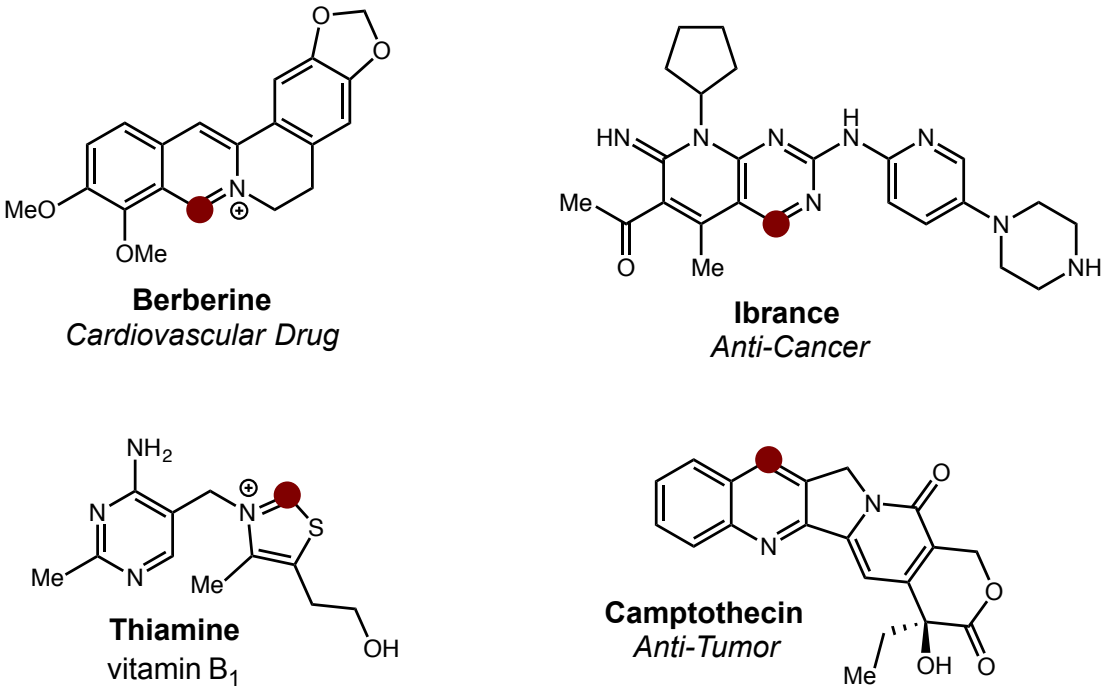
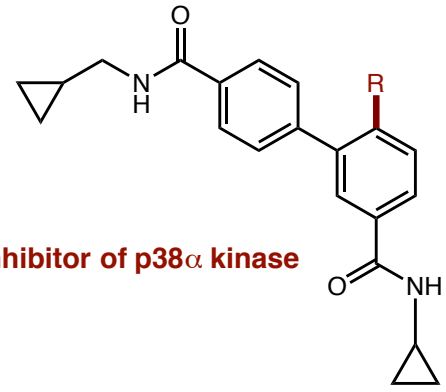


Figure 2 : Heteroarene-containing drugs

Functionalizations of drugs can improve their potency. One of the functionalizing methods is through post-synthetic alkylation. This type of functionalization for potency improvement is demonstrated as Methyl Effect[2]. Methylation is useful in optimizing properties of drug candidates that can significantly improve the IC_{50} (Figure 3).



R	IC_{50} (nM)
H	>16,000
Me	75

Figure 3 : Improvement of IC_{50} with methylation

One pathway to improve potency is to increase drug's biological activity[2]. The methylation provides alternative oxidation of the anti-inflammatory drugs (Figure 4). Through a normal pathway, the highlight thiozole group in Sudoxicam can be oxidized with enzyme CYP2C9. The pertinent decomposition resulted in the loss of its potency. Alternatively, the methylated Sudoxicam (Meloxicam) has a higher resistant. The pertinent product of oxidation is the alcohol that retains its potency as anti-inflammatory drug.

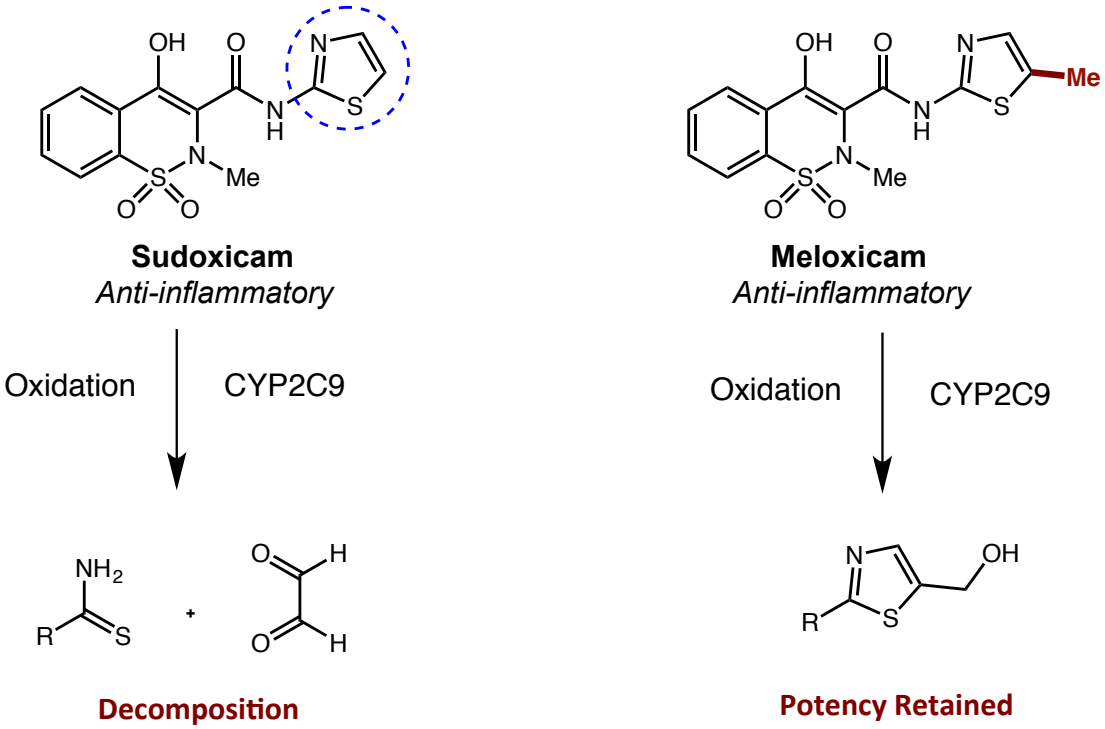


Figure 4 : Improved biological lifetime

The significant improvement on drug potency has drew interest to methylation/alkylation of pharmaceuticals. One of alkylation methods on these heteroarenes is Minisci reaction. This named reaction introduces alkyl groups to aromatic compounds, especially heteroarenes, via a radical substitution[3]. In the Minisci reaction of N-heteroarenes, the electron-deficient aromatic ring is

pronated on Nitrogen atom and becomes electrophilic readily for nucleophilically radical addition. The alklated product is formed after the addition of alkyl radial to aromatic ring, which is followed by further oxidation, elimination and rearomatization of the pertinent cation radical (Figure 5). The alkyl radical can be introduced by many means.

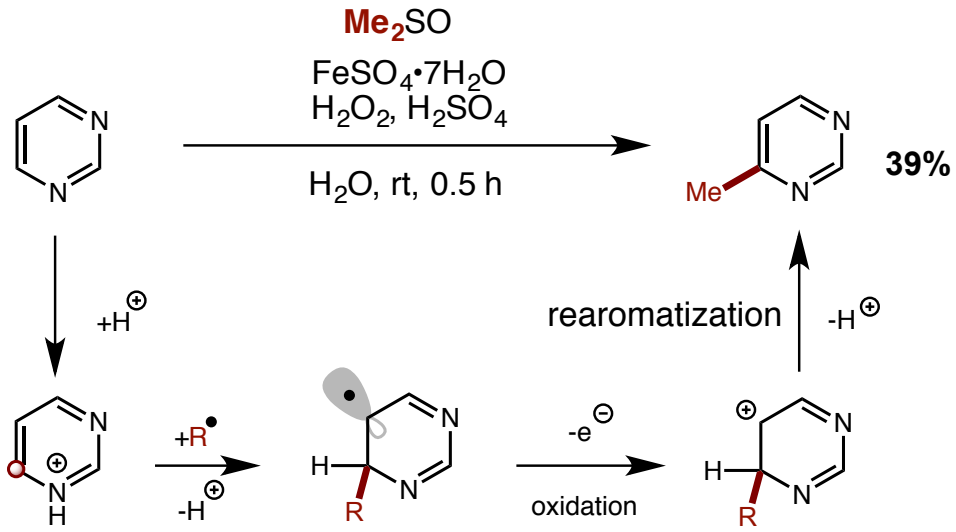


Figure 5 : Mechanism of Minisci reaction[4]

One of the pathways is the cleavage from dimethyl sulfoxide (DMSO) with Fenton's reagents (Figure 6). However, this condition is limited to methylation with the use of metal, which calls for a need for metal-free alkylation with more available alkylating sources.

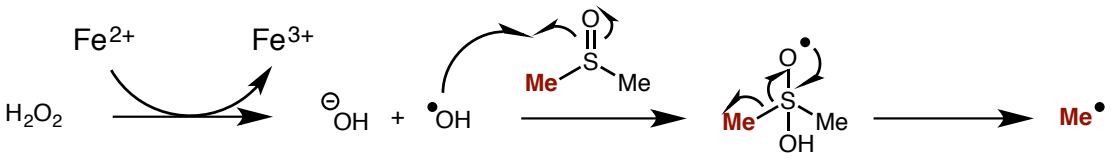


Figure 6 : Mechanism of alkyl radical formation

The interest of alcohols as alkylating sources is inspired by the Spin-Center Shift (SCS) mechanism in RNA deoxygenation[5] (Figure 7).

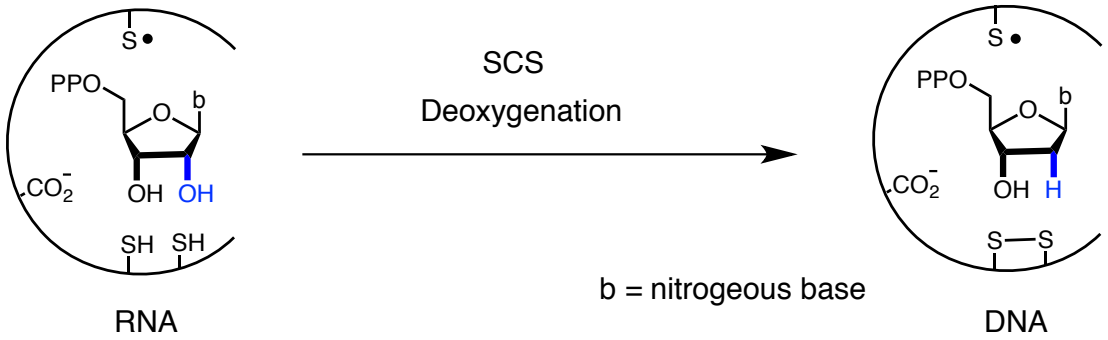


Figure 7 : Enzyme-catalyzed RNA deoxygenation

The alkyl radical is formed from deoxygenation of alcohols (Figure 8). In order to obtain the alkyl radical, the alcohol radical firstly formed after reduction or Hydrogen Atom Transfer (HAT). The removal of water introduces the carbocation. In order to stabilize this carbocation, the radical or the unpaired electron shifts to the positive charged carbon. The pertinent carbocation is stabilized by further elimination. The alkyl radical is formed via SCS pathway.

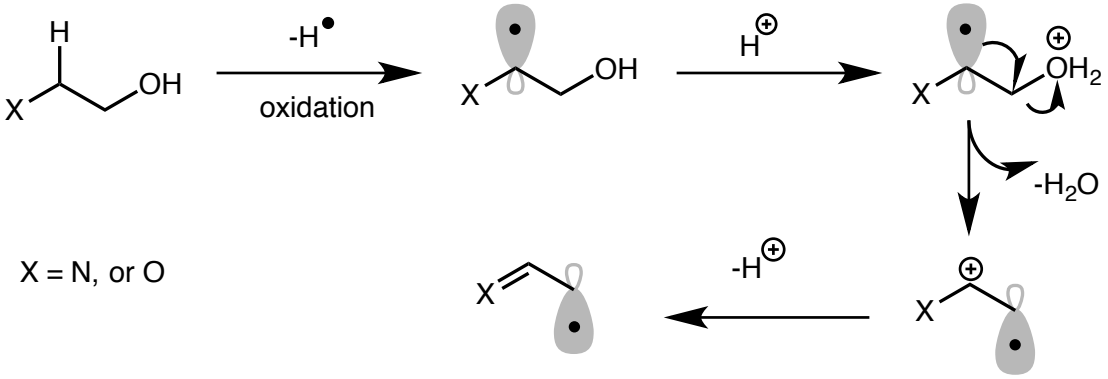


Figure 8 : Mechanism of alkyl radical formation via Spin-Center Shift

Compared with other alkylating resources, alcohols are nontoxic and readily available, and is able to form alkyl radial under metal-free condition. Thus, alcohols are competitive candidates for alkylation on pharmaceuticals and becomes the interest of investigation.

The goal of this project is to develop a novel alkylation of heteroarenes using readily available alcohols under metal-free condition (Figure 9). In this alkylation, the role of metal is replaced by heating, where oxidant radical is formed from the homolysis of pertinent peroxide. Different chemical conditions have been investigated as shown in Experimental Approaches.

Figure 9 : Hypothesis of metal-free alkylation with alcohols

The possible reaction pathways could be Minisci reaction and Spin-Center Shift alkylation (Figure 10). In the Minisci pathway, the early-stage oxidation by excess peroxide keeps the hydroxyl group on product. The removal of water before late-stage reduction introduces the alkylated product under SCS condition. The investigation has focused on the development and optimization of reaction conditions for SCS alkylation.

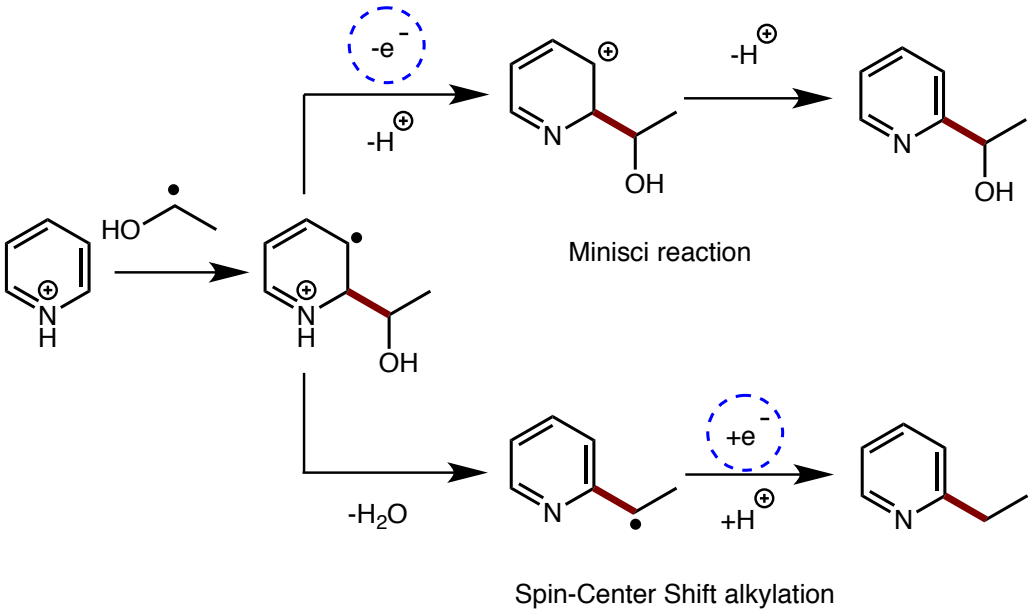


Figure 10 : Divergent additions of heteroarene

Experimental Approaches

The investigation of interest is the alkylation of an N-heteroarene derivative that is the biggest candidate for drugs. Successful alkylation of isoquinoline is observed via a peroxide-intermediated pathway by Jeremy Lear (Figure 11).

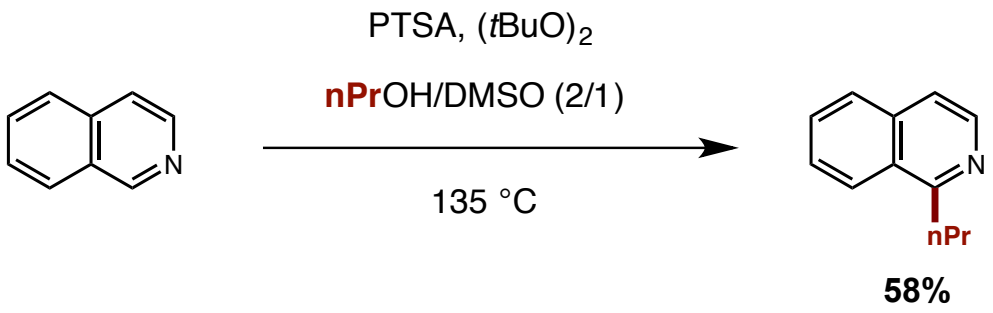


Figure 11 : Propylation of isoquinoline with n-propanol (with Jeremy)

In this alkylation, the radical from thermal hemolysis of peroxide serves as an initiator to reduce alcohol that introduces alcohol radical. The formed alcohol radical undergoes Minisci-type addition to the activated heteroarenes. In the

pertinent cation radical, α-hydrogen is transferred to nitrogen and this cation is neutralized with further elimination. An α-hydroxyl radical with SCS character is formed after Hydrogen Atom Transfer (HAT) and removal of hydrogen cation. Similar with alcohol radicals in RNA, this radical is deoxygenated via SCS pathway to introduce the new radical. At last, alkylated product is formed by propagating incoming alcohols. This propagation also provides new alcohol radicals for further SCS alkylation (Figure 12). This concept is called Spin-Center Shift alkylation or SCS alkylation for short.

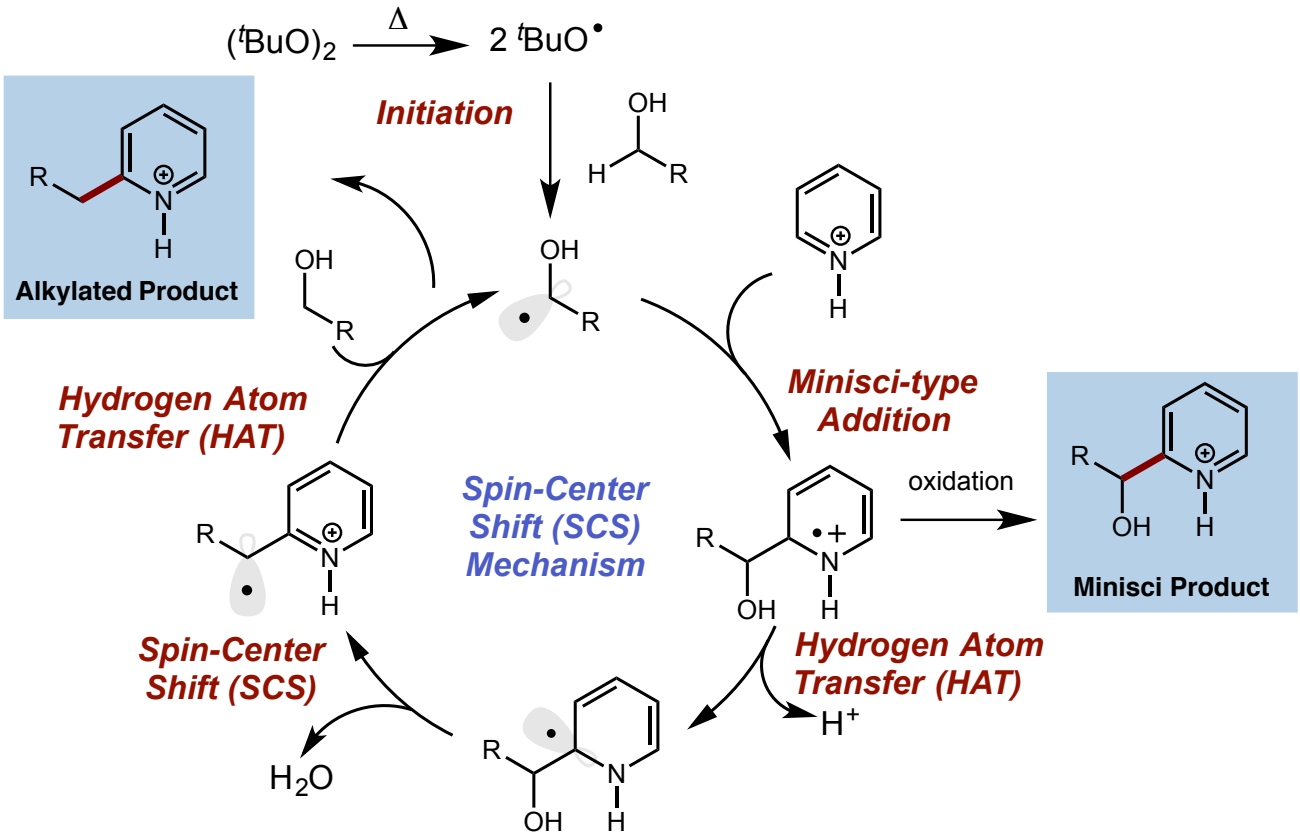
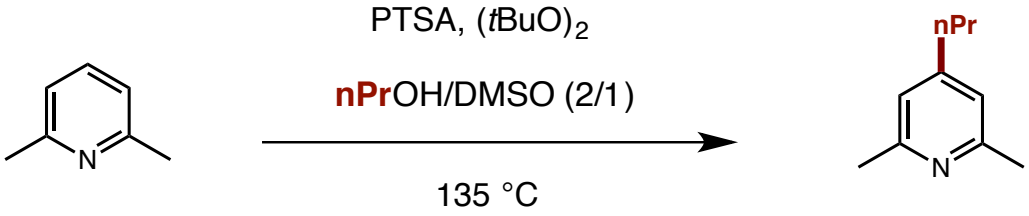


Figure 12 : Mechanism of SCS alkylation

In the heteroarene derivatives of commercial pharmaceuticals, the 2-positions are usually substituted and leaves 4-position available for alkylation, like Camptochecin (Figure 2). Based on this factor, 2,6-lutidine with 2-positions substituted is investigated with SCS alkylation (Table 1). In the reaction, p-toluenesulfonic (PTSA) acid was used to activate the 2,6-lutidine, tert-butyl peroxide as oxidant, dimethyl sulfoxide (DMSO) as solvent, and propanol as alkylating sources. The formation of 4-propyl-2,6-lutidine is observed with a trend that longer reaction period and more acidic condition provide higher yields. Based on the mechanism of Minisci reaction, the formation of methylated product is also expected due to the present of DMSO. However, no methylated product is observed. This result can b explained that the formation of alcohol radical is more favorable than the formation of methyl radical under the conducted condition, where the alcohol radical could be stabilized by its resonances.



Trial	(tBuO)2(eq.)	PTSA(eq.)	Time(h)	SM(%)	Target(%)
1	1.0	0.4	18	62.9	37.1
2	1.0	1.0	10	50.0	50.0
3	1.0	1.0	18	41.2	58.8

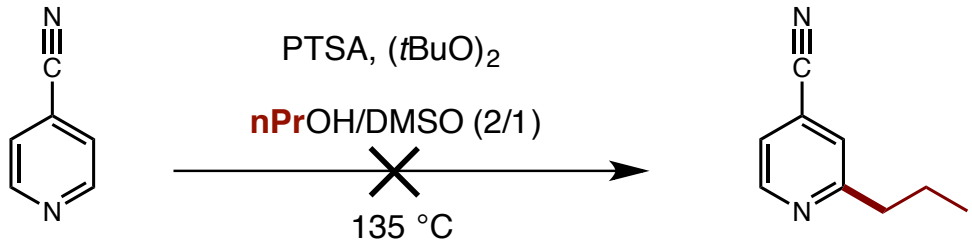
Table 1 : SCS alkylation of 2,6-lutidine

Based on the proposed mechanism (Figure 12) , one of the by-products is the Minisci-type product with hydroxyl group. In Minisci reaction, excessed peroxide is usually used to complete late-stage oxidation. The key of successful SCS alkylation instead of Minisci reaction is to use critical amount of peroxides. The expected peroxide can both initiate and direct reaction to a SCS alkylation pathway. From the experimental result, peroxide with one molar equivalent to substrate provides the highest yield of target. In addition, no Minisci-type product is observed in ^{1}H NMR spectrum.

In light of divergent reaction pathways, the product data of ^{13}C NMR and DEPT ^{13}C NMR are used to confirm the formation of 4-propyl-2,6-lutidine. The peak at 38 ppm is corresponding to the carbon connecting to aromatic ring, which carbon has two hydrogen atoms in SCS alkylated product and one hydrogen atom in Minisci-type products. In the spectrum of DEPT ^{13}C NMR, this peak is downward that indicates the present of CH_2 group. Thus, 2,6-lutidine is alkylated via SCS alkylation.

Motifs for followed investigation is the fact that heteroarenes are substituted by electron-withdrawing groups in many drug compounds. As a result, the aromatic ring becomes even more electron-deficient. An interest raised that whether SCS alkylation is able to alkylate these heteroarenes. 4-cyanopyridine was tested as electron poorer N-heteroarenes (Table 2). Acidity is tested as a control to optimize the reactivity of 4-cyanopyridine with SCS alkylation. However, no target molecule was observed. What is more, n-propyl isonicotinate

is formed and characterized under this condition. The cyanide group was hydrolyzed and esterified with n-propanol under acidic condition at high temperature.



Trial	(tBuO)2(eq.)	PTSA(eq.)	Time(h)	Yield(%)
1	1	0.2	7	0
2	1	0.2	22	0
3	1	0.4	7	0
4	1	0.6	7	0

Table 2 : SCS aslkylation of 4-cyanopyridine

Potency increase requires alkylation on specific position of heteroarene derivatives that calls for a need of selectivity in alkylation. The acid-activated SCS alkylation gives less control on the selectivity. In order to selectively alkylate heteroarenes, new activation methods are being investigated. meta-Chloroperoxybenzoic acid (mCPBA) is an efficient oxidant that activating 2-positions. The activated 2-position become more electrophilic and is ready for nucleophilic radical substitution. As a result, the formation of 2-Alkyated product is favorable over 4-Alkylated product, where the selectivity is established. Furthermore, in order to obtain 4-Alkylated product, N-alkyl activation is introduced for N-heteroarenes. Alkylation on nitrogen can activate the aromatic

ring and also contribute to selectivity in alkylation (Figure 13). The bulky alkyl group, like isopropyl group, on nitrogen increases steric hindrance on 2-positions, resulting a major formation of 4-Alkyalted product (Figure 14). The 2- or 4-alkylated products are formed after further removal of this activating group.

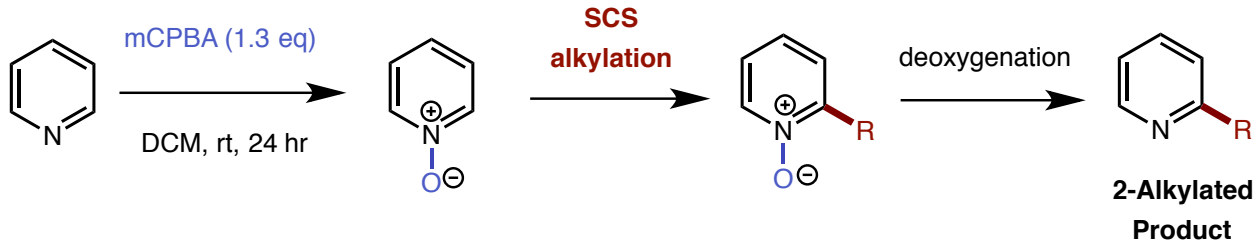


Figure 13 : Proposal for selectivity of 2-alkylated product

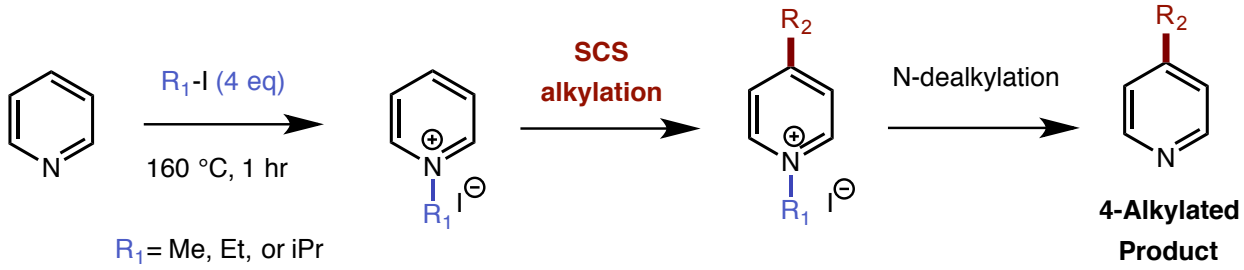


Figure 14 : Proposal for selectivity of 4-alkylated product

The alternative activations were investigated on 2,6-lutidine. Formation of N-oxide (Figure 15) was observed. In order to complete activating and deactivating circle, basic deoxygenation[6] (Figure 16) was tested on N-oxide alkyl 2,6-lutidine (Figure 17).

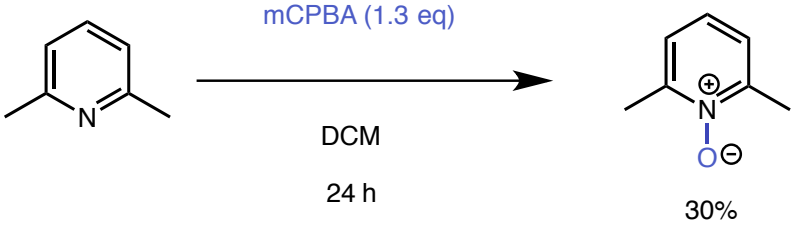


Figure 15 : N-Oxidation with mCPBA

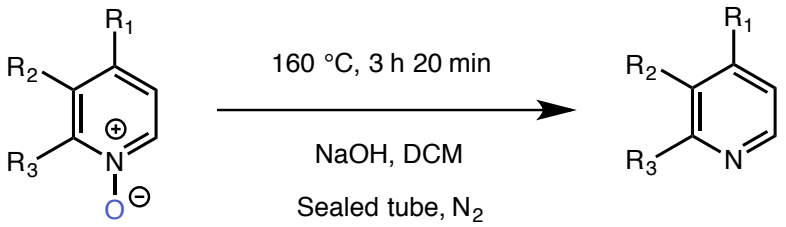


Figure 16 : Deoxygenation under basic conditions

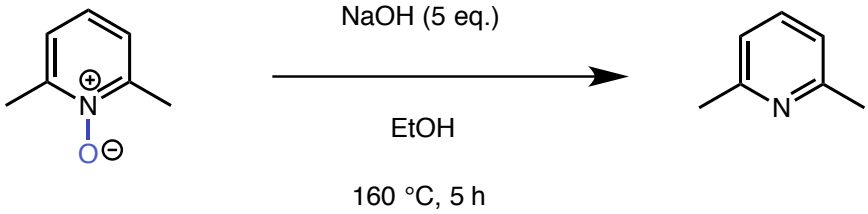
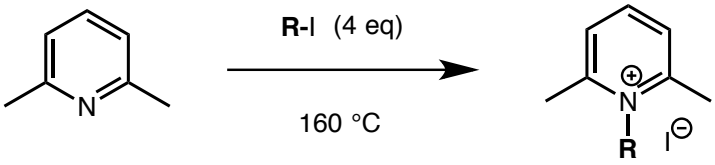


Figure 17 : Base-mediated deoxygenation of N-oxide

However, only N-oxide was observed by ^{1}H NMR spectrum and Thin Layer Chromatography (TLC) plate analysis. Other combinations of base and solvent will be used for further investigation. Formation of N-alkyl lutidinium (Table 3) was observed.



Trial	R	Time (h)	Yield(%)
1	isopropyl	13	0
2	isopropyl	44	0
3	ethyl	13	80

Table 3 : N-alkylation with alkane iodide

Further removal of activating group or N-dealkylation is inspired by the Hoffman elimination[7] (Figure 18). The iodide anion is removed by silver cation and followed by base-assisted cleavage of the C-N bond.

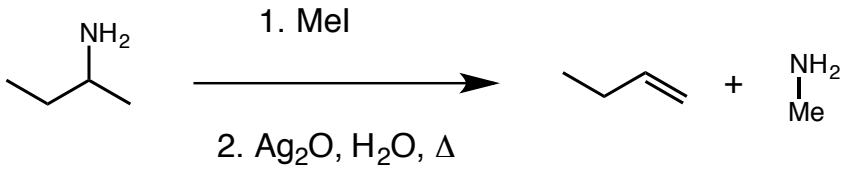


Figure 18 : Hoffman elimination

Summary

SCS method provides a metal-free alkylation of heteroarenes using alcohols. Our recent investigations have demonstrated successful alkylation of isoquinoline, quinoline, 2,6-lutidine and 4-*t*Butylpyridine (Figure 19). These studies were carried out with Jeremy Lear and Ani Mustafa. Alternative activation of heteroarenes with selectivity is being investigated (Figure 20).

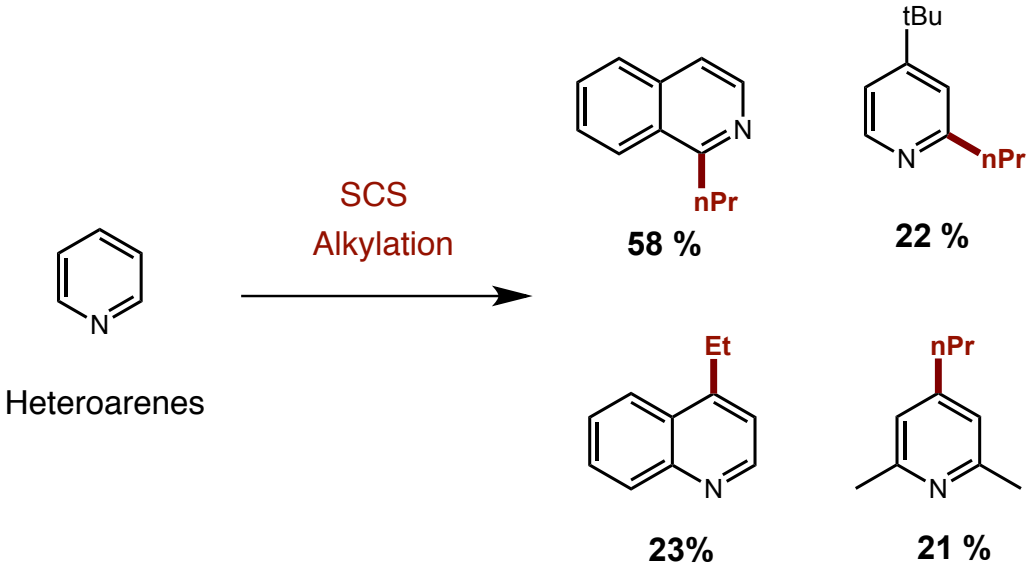


Figure 19 : Substrate scope of successful SCS alkylation (with Jeremy and Ani)

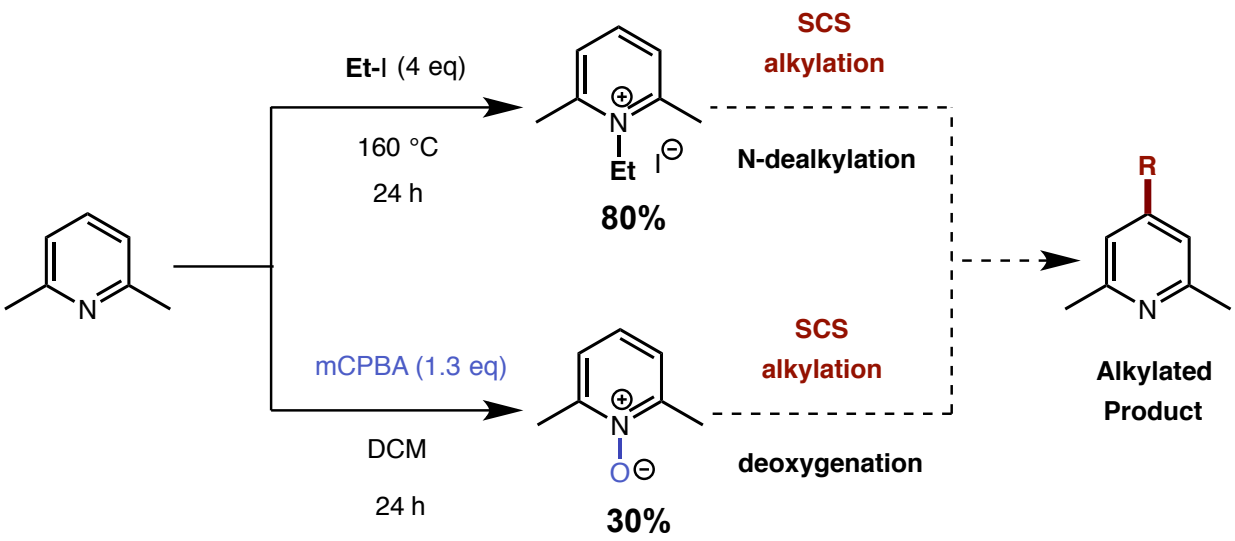


Figure 20 : Alternative activation of SCS alkylation

Future Investigation

A library of pharmaceutical derivatives will be further interests of SCS alkylation. Efficient removal of alternative activating group will be also a focus in this project. Modified SCS alkylation provides a new pathway to alkylated pharmaceutical derivatives with the promise of improvement in potency of these pharmaceuticals.

References

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Supporting Information

General Procedure: A solution of 2,6-lutidine (50 mg, 0.47 mmol), p-Toluenesulfonic acid (0.0925 g, 0.19 mmol), DMSO (0.5 mL) and nPrOH (1.0 ml, 13.36 mmol) were mixed in a 8-mL screw thread vial and degased via Freeze-Pump-Thaw. $(tBuO)_2$ was added last to the mixture under purging with N_2. The reaction vial than was transferred to a pre-heated hot plate and ran for 8 hours at 135 °C.

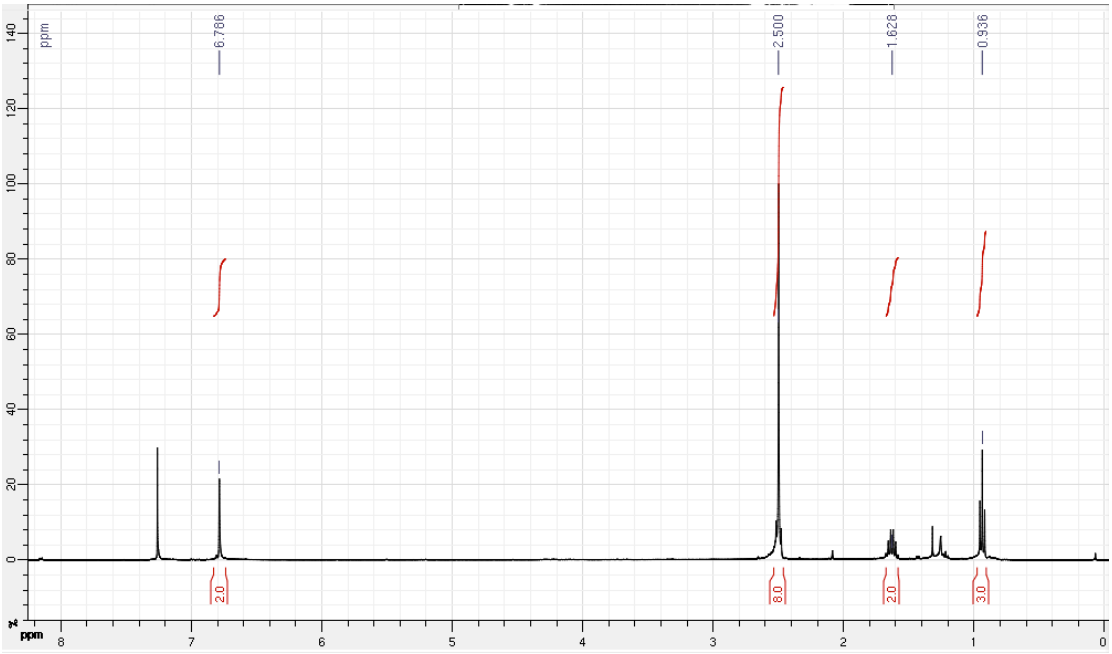
General Work-Up: Sodium bicarbonate was used to neutralized the reaction mixture to a pH of 8 – 9. To a separatory funnel, 10 mL of distilled water was added. Dichloromethane (3 × 10 mL) was used to wash the reaction mixture to extract product from aqueous layer into organic layer. The alkylated product was isolated through column.

4-propyl-2,6-lutidine

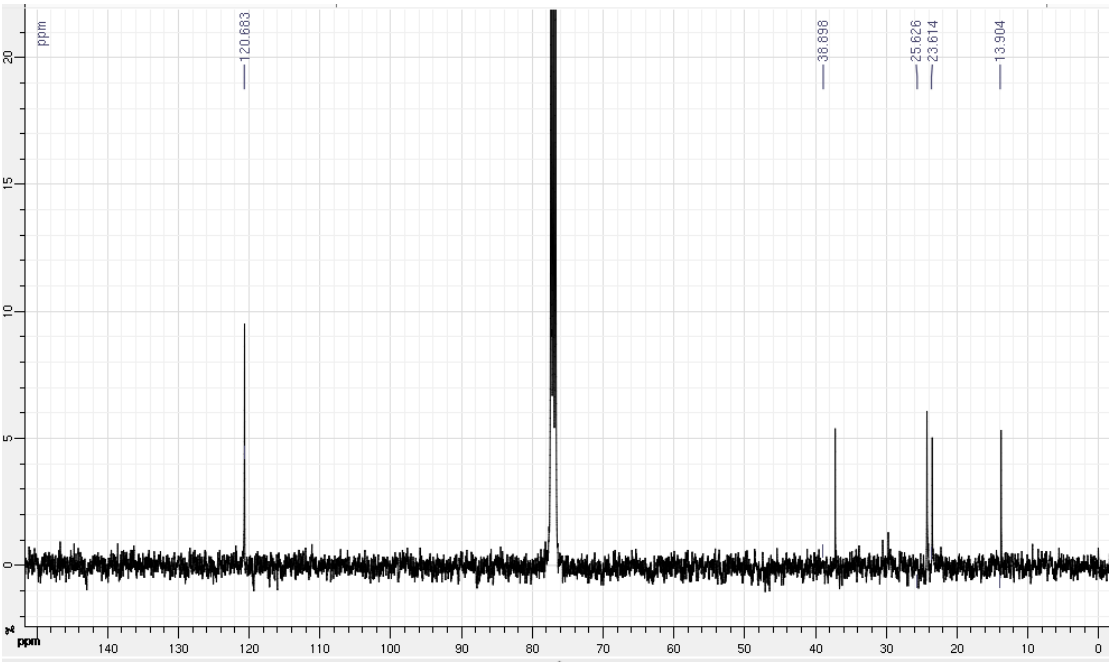
^{1}H NMR (400 MHz, CDCl3) δ 6.906 (s, 2H), 2.50 (s, 6H and t, 2H), 1.62 (m, 2H), 0.94 (t, 3H) ppm; **^{13}C NMR** (400 MHz, CDCl3) δ 120.63, 38.90, 25.63, 23.61, 13.90 ppm. **MS:** Expected (m/z 149.1); Observed (m/z 151.0); **IR**: 2960.38, 2923.28, 2852.97, 1637.121609.49, 1567.88 cm^{-1}

***n*-Propyl Isonicotinate**

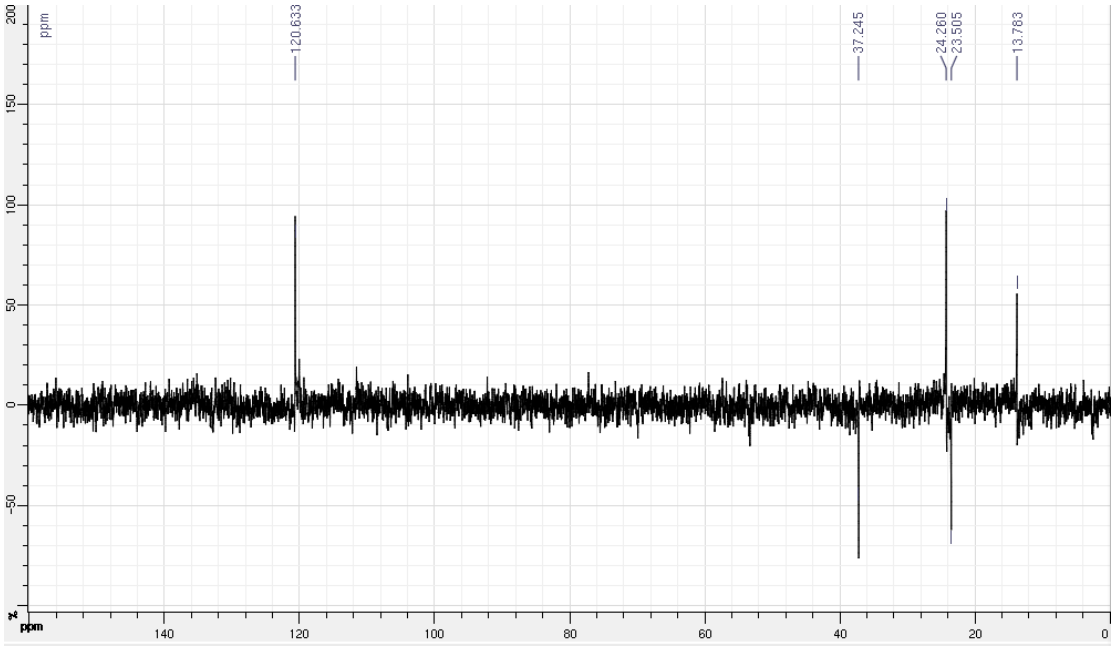
^{1}H NMR (400 MHz, CDCl3) δ 8.77 (m, 2H), 7.84 (m, 2H), 4.31 (t, 2H), 1.80 (m, 2H), 1.03 (t, 3H). **^{13}C NMR** (400 MHz, CDCl3) δ 165.73, 150.59, 137.61, 122.84, 67.35, 22.00, 10.44 ppm.



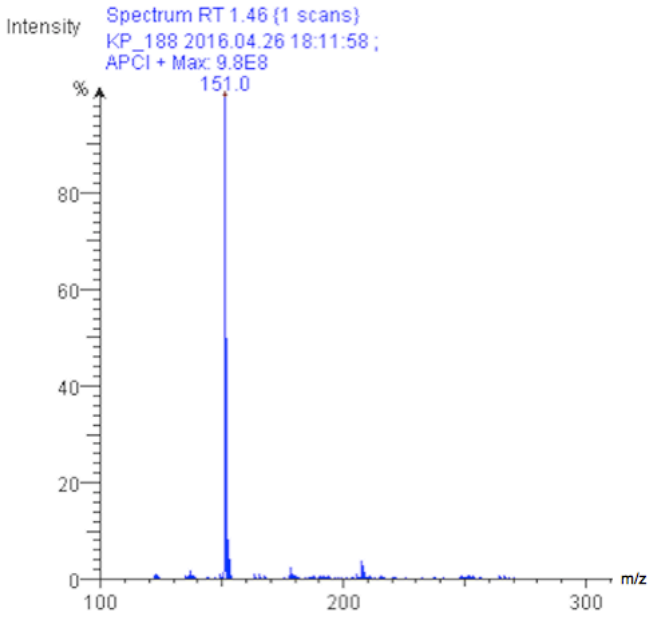
^{1}H NMR spectrum of **4-propyl-2,6-lutidine**



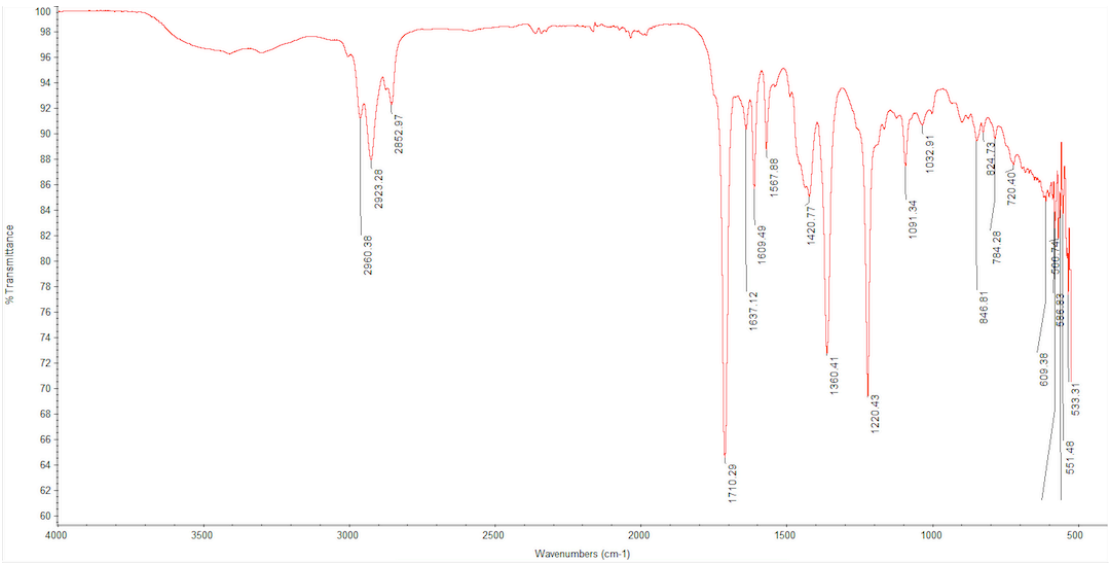
^{13}C NMR spectrum of **4-propyl-2,6-lutidine**



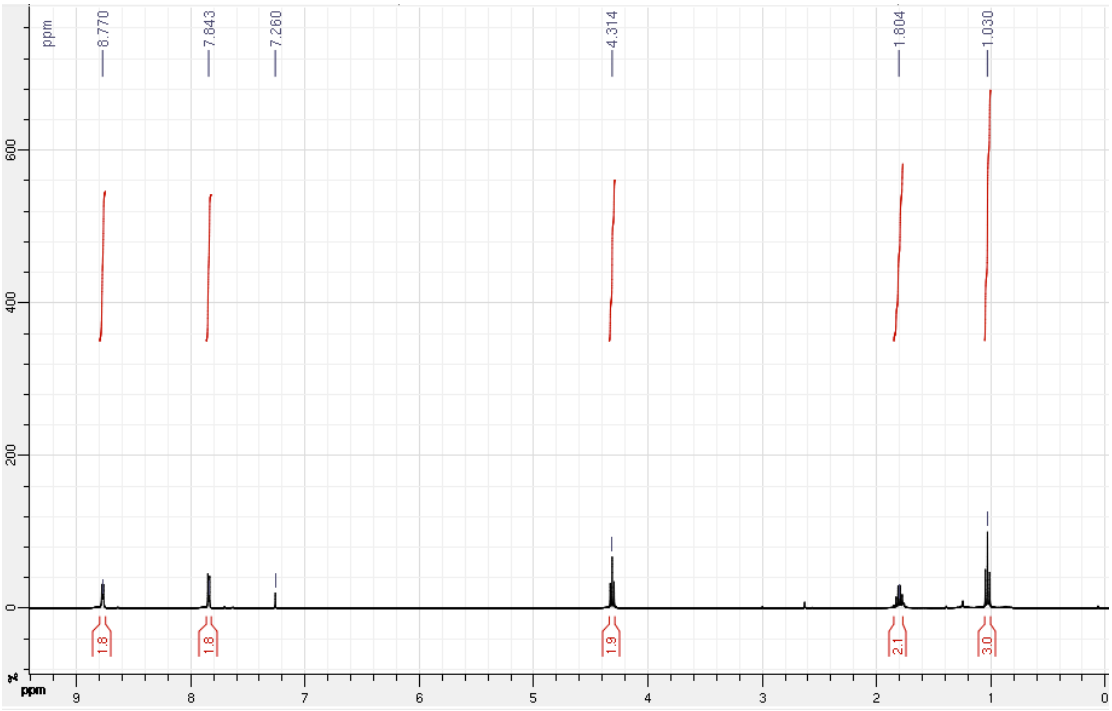
DEPT ^{13}C NMR spectrum of **4-propyl-2,6-lutidine**



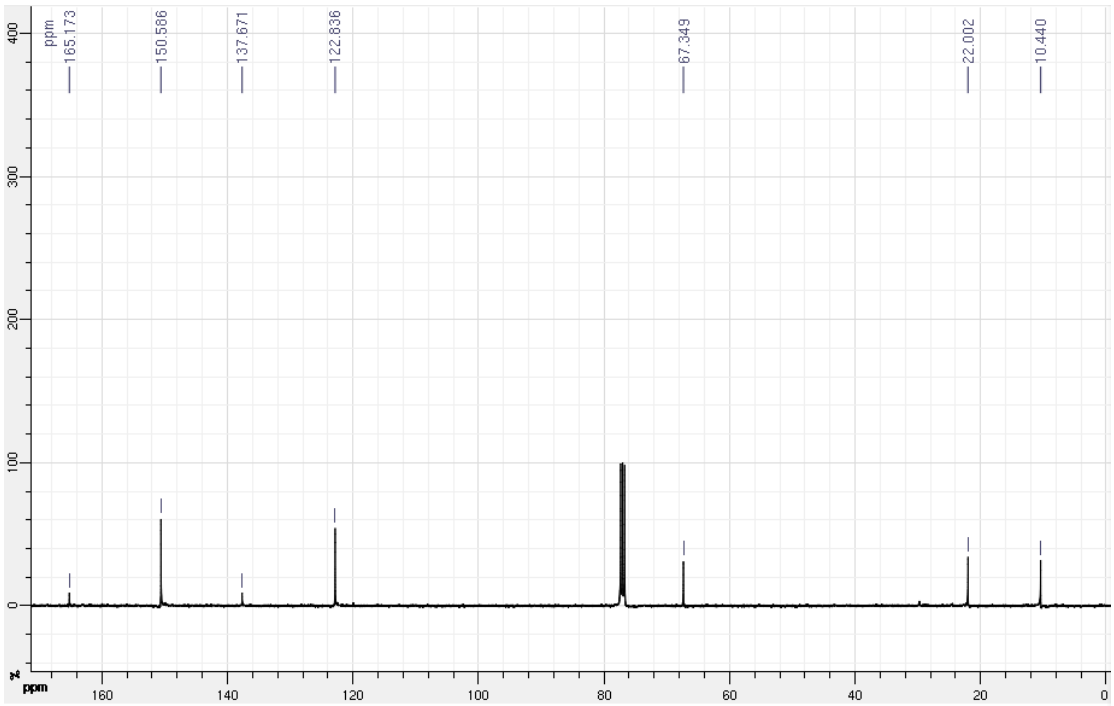
Mass spectrum of **4-propyl-2,6-lutidine**



IR spectrum of **4-propyl-2,6-lutidine**



^{1}H NMR spectrum of **n-Propyl Isonicotinate**



^{13}C NMR spectrum of **n-Propyl Isonicotinate**